BIO FAX!

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# Brown Out

## A Student-Designed Scientific Method Experiment

### Introduction

Ever bite into an apple and hit a brown spot? Did you spit the brown out? What causes apples, potatoes, pears, and other foods to turn brown? Can it be prevented?

### Concepts

- Enzyme activity
- Scientific method

### Materials

Apples

Student experimental designs will dictate equipment needs.

### *Safety Precautions*

*Follow all normal laboratory safety precautions. Follow safe lab procedures with all the equipment used by students in their experimental designs. Do not allow students to work alone or conduct unsafe experiments at home. Please review current Material Safety Data Sheets for all chemicals that will be used in the lab.*

### Preparation

This lab will be introduced on day one with a teacher-led demonstration followed by time for discussion and designing experiments. The next lab period should be spent conducting the student-designed experiments.

On day one, provide each team with a slice of cooked apple, a slice of raw apple that has been sitting out for at least three hours, and a slice that has just been cut. (Prepare cooked apples by boiling, baking or microwaving whole apples.)

Ask each team to generate a list of differences they can observe among the apple slices and then to consider these questions:

1. In what order were the slices most likely cut?
2. Which slices are similar in color?
3. How uniform is the color?
4. How deep is the color in the apple?
5. How can this browning be prevented?

After an initial observation period, have each team list their ideas on their record sheet. Give students time to think. Then have a discussion to pool all of their ideas regarding how the browning can be prevented. Ideas will likely come from personal experiences with apples. Some will recall cooking experiences or experiences with plastic wraps, lemon juice, water or oils. Eventually consider the fact that the browning phenomenon is a chemical reaction that is controlled by an enzyme (if you have previously studied enzyme activity). Ask students to consider all of the variables that they predict may affect this chemical reaction. The lists will be varied but might include light, air, pH, temperature, moisture, carbon dioxide, and oxygen.

Next, give each team time to select a hypothesis and design an experiment to test their hypothesis. Ask probing questions of each group encouraging them to quantify and control their experiment as much as possible. Try to direct students toward designs that are possible in your lab and with readily available materials. Also remind teams that experiments that disprove a factor are just as valid as experiments that prove a factor. Have each team submit a materials request list before leaving class. This will give you time to locate the needed materials before the next class period.

Be sure to approve each experiment before students start their work. Give students guidelines for the project. Possible guidelines are provided at the end of this Bio-Fax!

## Procedure

1. Consider the demonstration apples provided by your teacher.
2. Record all the class discussion ideas about why apples turn brown and how to prevent the browning.
3. Which hypothesis has your team selected? How do you anticipate stopping the browning?
4. Describe your experimental design in detail. Be sure to include:
   *a.* Materials list—all the materials needed for the experiment. Be sure to note which materials you need to collect yourself.
   *b.* Detailed description of your procedures and experimental setup. (Use drawings if they are helpful.)
5. Conduct your apple experiment.
6. Make appropriate data tables and record the results from your experiment.
7. Write up your complete experiment. Include all the information from above. Discuss any unusual results or discrepancies as they relate to your original hypothesis.
8. Continue work with additional experiments or literature research.

## Disposal

Please consult your current *Flinn Scientific Catalog/Reference Manual* for general guidelines and specific procedures governing the disposal of laboratory waste. It is anticipated that all materials used in this experiment will be safe.

## Tips

- Do this activity after your standard lab work with enzymes and when students are ready to design and conduct their own experiments.
- Establish a procedure to approve students' experiments before they begin their experiments. Encourage students to incorporate controls into their experiments and to design experiments that are within the time and equipment constraints you may have. Do not stifle creative ideas for experiments, however, even if it is not possible to perform them in class.
- Some possible ideas for experiments:

  A. Oil the slices.

  B. Place apple slices under water.

  C. Use different wrapping materials. (*Example:* Does the wrap let oxygen through? Saran™ Wrap vs. Glad® Wrap).

  D. Generate oxygen to speed up the browning process.

  E. Generate carbon dioxide and test its effect.

  F. Change the pH (lemon, vinegar, acid, base, etc.) of the apple slices.

  G. Investigate the effect of temperature.

  H. Use preservatives such as salt, citric acid, ascorbic acid (Vitamin C), sugar, etc.
- You may want to avoid duplicate experiments. Instead, have all groups work on different variables and then pool the results as a post-lab discussion.



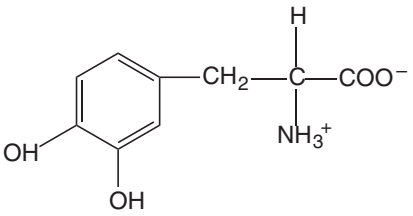
## Discussion

The "browning phenomenon" exhibited by some fruits and vegetables is explained in terms of the basic chemistry of naturally occurring phenolic compounds and enzymes. Phenols such as dihydroxyphenylalanine (see Figure 1) are colorless compounds. They are, however, easily oxidized and the oxidation products are often colored. The oxidation reactions are generally controlled by enzymes and factors affecting the enzymatic reactions can influence the rate of oxidation and therefore the color change.

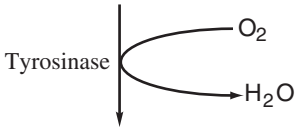
In this experiment, the reactions that cause the browning of apples are the same type of reactions involved in the production of melanin (brown pigment) in our skin. The key part of the reaction mechanism is summarized in Figure 1.

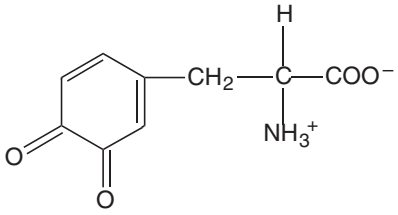
The resulting colored products are similar to those that increase in amount in the human skin when exposed to sunlight. Exactly how the browning reaction might benefit the apple (if at all) is not clear.

Not all fruit cells contain "dopa-like" compounds in their cells and, therefore, not all fruits will turn brown when damaged. It is the combination of the enzyme in the presence of oxygen that causes the reaction. Cutting the fruit disrupts the cells, releases their content, and exposes them to the oxygen in the air. (Normally the apple skin, waxed coatings, etc. prevent oxygen from getting into the fruit and turning it brown.) When cut, the fruit will only turn brown to the depth which oxygen will diffuse. Thus, often only the surface layer is brown and lower depths (with no oxygen penetration) will remain colorless.




Dihydroxphenylalanine
(dopa)




Phenylalanine-3, 4-quinone
(dopaquinone)

**Figure 1.** Oxidation reaction

Preventing browning can be accomplished by either destroying the enzyme (cooking for example) or by preventing the cut surface from being exposed to oxygen (keeping under oil, water, cover, etc.) Precut apple slices sold in grocery stores are treated with ascorbic acid (Vitamin C), a naturally occurring antioxidant that prevents oxidation of other substances. Any other factors affecting the rate of the enzyme activity will also affect the rate of browning. Students may wonder about the cause of browning in uncut fruit. Naturally occurring air spaces, bruising, insects, and other physical damage may explain the browning of uncut fruit. Waxing the skin of apples may not only prevent drying, but by cutting off the oxygen, the wax will help prevent any chemical reactions requiring oxygen.

## Connecting to the National Standards

This laboratory activity relates to the following National Science Education Standards (1996):

***Unifying Concepts and Processes: Grades K–12***
- Evidence, models, and explanation
- Constancy, change, and measurement

***Content Standards: Grades 5–8***
- Content Standard A: Science as Inquiry
- Content Standard C: Life Science, structure and function in living systems
- Content Standard G: History and Nature of Science

***Content Standards: Grades 9–12***
- Content Standard A: Science as Inquiry
- Content Standard C: Life Science, matter, energy, and organization in living systems
- Content Standard G: History and Nature of Science, nature of scientific knowledge

## References

Markstein, J. A.; Posner, H. B. *Student-Directed Investigations in Enzymology for Introductory College Biology, The American Biology Teacher,* Volume 60, No. 1, Reston, VA, January 1998.

Zubay, G. *Biochemistry,* 3rd ed.; Wm. C. Brown: Dubuque, IA, 1993.

